Thermal Death Points of Pathogenic Bacteria in Cream and Ice Cream*

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CONSIDERABLE study has been made of the thermal death points of bacteria in milk. Little has been done to determine whether the pasteurization temperature and time (143.5° F. for 30 minutes)[1] recommended is sufficient to kill the non-spore bearing pathogenic bacteria which may contaminate cream or ice cream. The question arose as to whether the higher percentage of butter fat in cream or ice cream would act to protect the bacteria in ordinary pasteurization.

The methods used by Park[2] formed the basis for the test procedures. The organisms used were:

1. Two strains of *B. typhosus* recently isolated from cases
2. Two strains of beta type hemolytic streptococci, one from scarlet fever, the other from septic sore throat
3. A culture of tubercle bacillus of the bovine type

The typhoid bacilli and the streptococci were grown in veal broth for 18 hours, then diluted with sterile broth to an appropriate density. The tubercle bacilli were cultivated on glycerine beef broth. The pellicle, after 3 weeks' growth, was filtered off, weighed and emulsified in sterile physiological saline, and diluted to a final concentration of 0.2 mg. of dry tubercle bacilli per c.c. of suspension.

Cream with 50 per cent butter fat content was sterilized in the autoclave for 30 minutes at 15 lb. pressure. For the test, 9.9 c.c. were measured into each of a series of small bottles. After heating until the cream was at the test temperature, 0.1 c.c. of the culture was added to each bottle. These were then stoppered tightly, vigorously shaken, and submerged in the water bath at the test temperature. Five temperatures from 135° to 145° F. were used. Samples were taken at various intervals. After heating for the desired time, the bottles were removed from the bath, shaken, and covered with cracked ice. After chilling, the cream was tested as follows:

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1. For the isolation of *B. typhosus,* six brilliant green agar plates were used. Two were inoculated with 0.2 c.c. each and one with 0.1 c.c. of the cream, this inoculum being streaked over the other three plates. At the same time, two tubes of bile peptone solution were inoculated with 2 c.c. of the cream. These were incubated for 18 hours for enrichment, and the bile was then plated on brilliant green agar. All plates were examined after 18 hours' incubation and slide agglutinations done with all suspicious colonies. As a further check on positive findings, the suspicious colonies were fished to Krumwiede's triple sugar medium.

2. For isolation of the streptococci, 1 c.c. and 0.1 c.c. amounts were pipetted into tubes of melted blood agar, which, after mixing, were poured into Petri dishes. One c.c. of each sample was also inoculated into tubes of blood broth for enrichment, and after 24 hours' incubation, plated with blood agar. All hemolytic colonies appearing on these two series of plates were fished into blood broth and incubated. As a final check, blood plates were poured from all fishings to determine that the streptococci recovered were of the beta hemolytic type.

3. Guinea pigs were used to test the survival of tubercle bacilli, three pigs for each sample. Each pig was inoculated subcutaneously into the knee-fold region with 1 c.c. of the cream. Pigs dying within 8 days were discarded; those dying at a later date were autopsied; those surviving 42 days were killed and autopsied. Smears were made of all suspicious lesions and examined for acid-fast bacilli. When no acid-fast bacilli were found, subinoculations were made into other guinea pigs.

The results obtained in the above series of tests are given in Table I. The streptococci did not survive heating in cream at 135° F. for 5 minutes, nor at 140° to 142° F. for 1 minute. At 143.5° and 145°

TABLE I

THERMAL RESISTANCE OF BACTERIA IN CREAM

		Survival of Organisms after:								
Organism	Temperature Fahrenheit	½ min.	1 min.	2 min.	3 min.	4 min.	5 min.	7 min.	10 min.	20 min
B. typhosus	135.0	+	+	+	+	+	+	+	—	—
	140.0	+	+	+	+	+	—	—	—	—
	142.0				*—	—	—	—	—	—
	143.5	+	+	—	—	—	—	—	—	—
	145.0	+	+	—	—	—	—	—	—	—
Streptococci	135.0	+	+	+	+	+	—	—	—	—
	140.0	+	—	—	—	—	—	—	—	—
	142.0	+	—	—	—	—	—	—	—	—
	143.5	+	†	—	—	—	—	—	—	—
	145.0	+	†	—	—	—	—	—	—	—
B. tuberculosis	135.0									*—
	140.0	+	†	+	+	+	+	+	—	—
	142.0	+	†	+	+	+	+	—	—	—
	143.5	+	†	+	+	+	+	—	—	—
	145.0				*—	—	—	—	—	—

* Shortest time tried.

† No 1 minute specimen tried.

Unheated controls—original numbers of organisms per c.c.: *B. typhosus,* 87,000; Streptococci, 90,000; *B. tuberculosis,* 6 pigs inoculated developed generalized tuberculosis.

Average life of control pigs was 33 days. None survived full test time of 42 days.

+ indicates the organisms survived.

– indicates the organisms did not survive.

F., no test was made after 1 minute, but after 2 minutes there was no survival of streptococci. Typhoid bacilli heated at 135° F. were killed in less than 10 minutes, at 140° in less than 5 minutes and at 142° to 145° F. in less than 3 minutes. Tubercle bacilli in cream were killed * in less than 20 minutes when heated at 135°, in less than 10 minutes at 140°, in less than 7 minutes at 142° to 143.5°, and in less than 3 minutes at 145° F. In every case, controls of unheated infected cream contained a much larger number of pathogenic bacteria than cream would contain under natural conditions of contamination.

Similar experiments were done using an ice cream mixture supplied by one of the local ice cream manufacturers. This consisted of cream, milk, condensed milk and sugar in the proportions used commercially. The resistance of *B. typhosus,* streptococci, tubercle bacilli and a strain of *B. diphtheriae* was tested.

The technic for the determination of the survival of the *B. typhosus,* streptococci, and tubercle bacilli, was the same as in the preceding experiment. For the isolation of *B. diphtheriae* 1 c.c. and 0.1 c.c. amounts of the samples were pipetted into tubes of melted serum agar.

TABLE II

THERMAL RESISTANCE OF BACTERIA IN ICE CREAM MIXTURE

Organism	Temperature Fahrenheit	Survival of Organisms after: ½ min.	1 min.	3 min.	5 min.	6 min.
B. typhosus	145	+	+	+	—	—
	150	+	+	+	—	—
Streptococci	145	+	+	+	—	—
	150	+	+	+	—	—
B. diphtheriae	145	—	—	—	—	—
	150	—	—	—	—	—
B. tuberculosis	145†	+	+	+	+	—
	150†	+	+	—	—	—

Numbers of organisms in unheated controls:

B. typhosus ..... 87,000 per c.c.
Streptococci ..... 46,000 per c.c.
B. diphtheriae ..... 2,000 per c.c.
B. tuberculosis ..... 6 pigs inoculated all developed generalized tuberculosis *

* One of the control pigs survived 42 days. Average life of others was 34 days.
† All pigs showing generalized tuberculosis survived 42 days.
+ indicates organisms survived.
– indicates organisms did not survive.

After mixing, the agar was poured into Petri dishes. One c.c. of each sample was also measured into each tube of serum broth for enrichment. After 18 hours' incubation both plates and tubes were examined for the presence of *B. diphtheriae.*

The results of these tests are shown in Table II.

B. diphtheriae in an ice cream mix did not survive heating for ½

* Failure to produce infection in guinea pigs is considered as meaning bacilli were killed. These figures are somewhat lower than frequently given.

minutes at 145° to 150° F. *B. typhosus* and the streptococci were killed in less than 5 minutes at these temperatures. Tubercle bacilli were killed within 6 minutes at 145° F., and within 3 minutes at 150° F. In comparing these results with those of the cream experiments, a slightly greater resistance was noted in the mix. This, however, was so slight that it falls within the limit of technical variation.

CONCLUSION

The time and temperature (30 minutes at 143.5° F.) recommended by the Committee on Dairy Products and Eggs, American Public Health Association, for the pasteurization of milk allows an ample margin of safety for the pasteurization of cream and of commercial ice cream mix.

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REFERENCES

1. Report of Committee on Dairy Products and Eggs (unpublished), A. P. H. A., Oct. 17, 1928.
2. Park, William H. Thermal Death Points of Pathogenic Bacteria in Milk, *A. J. P. H.*, 17, 1: 36 (Jan.), 1927.

National Children's Bureau of Argentina

A BILL for the establishment of a National Children's Bureau (Direccion nacional de la ninez) was recently introduced into the Congress of Argentina.

The main functions of the bureau will be to study the problems of improving the race of the future, to organize local councils in the capitals of the provinces and territories, and to establish institutions necessary for the carrying out of the work. The local councils are to open dispensaries for the treatment of physical and mental disorders of children. The bureau is to coöperate with the juvenile courts on the principle that a delinquent child is a sick child and must be treated as such, and is to establish reformatories with facilities for the medical treatment of the inmates.

The bill also provides for the establishment of a medical register of all school children in the country; this register to consist of individual records, each prepared after a thorough physical examination and mental test.—*Boletin del Museo Social Argentino,* Buenos Aires, Feb., 1930, p. 96.